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Original article

GENETIC POLYMORPHISMS OF CYP11B2 GENE IN HYPERTENSION

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Abstract: *Hypertension is a complex, multifactorial disease influenced by both genetic and environmental factors. Essential hypertension is partially hereditary, with alterations in aldosterone synthesis and regulation potentially playing a role. Low-renin hypertension, characterized by an elevated ratio of aldosterone to plasma renin activity, is associated with inappropriate aldosterone biosynthesis. The cytochrome P450 family 11 subfamily B member 2 (CYP11B2) gene, which encodes aldosterone synthase, has been suggested as a possible candidate in the genetic predisposition to hypertension.* ***Objective:*** *This study aimed to investigate the association between the CYP11B2 gene polymorphism −344T/C (HaeIII) and hypertension by analyzing the allelic frequency and genotype distribution in hypertensive patients and healthy controls.* ***Methods:*** *A total of 65 hypertensive cases and 42 controls (including both males and females) were enrolled. Genomic DNA was extracted, and the CYP11B2 −344T/C polymorphism was analyzed using PCR-RFLP (Polymerase Chain Reaction-Restriction Fragment Length Polymorphism). The allelic frequency and the distribution of TT, TC, and CC genotypes in both cases and controls were examined.* ***Results:*** *The analysis revealed that the control group exhibited a higher frequency of the TT genotype, while the patient group displayed an increased prevalence of both CT and CC genotypes. Notably, the C allele was not observed in the control samples but was present at a significantly higher frequency in hypertensive patients. Statistical analysis indicated that the C allele was a significant risk factor for hypertension ($p < 0.01$).* ***Conclusion:*** *Our findings suggest that the CYP11B2 −344T/C polymorphism may contribute to the genetic predisposition to hypertension, particularly in individuals with a family history of the disease. These results highlight the importance of genetic factors in hypertension susceptibility and underscore the need for follow-up studies to assess the long-term risk and incidence of hypertension in individuals carrying genetic risk factors.*

Keywords: Hypertension, CYP11B2, Polymorphism, PCR-RFLP, Case, Control

Introduction

Hypertension is a multi-factorial disease that influences both environmental and genetic components. From 20% to 40% of the blood pressure variation in the general population can be characterised by genetic factors (1). Low-renin hypertension is described by a high ratio of aldosterone to plasma renin activity, which may result in inappropriately increased aldosterone biosynthesis. About 10% to 20% of patients with essential hypertension exhibit high ALD/PRA (2).

Aldosterone is the principal mineralocorticoid hormone, Sodium balance and volume homeostasis are regulated by Aldosterone. Several common polymorphisms associated with hypertension have been described in the CYP11B2 gene. Polymorphisms may affect the synthesis of aldosterone or its regulation may have effects on blood pressure (3). Aldosterone is produced exclusively in the adrenal zonaglomerulosa, and its secretion is regulated primarily by serum levels of angiotensin II (ANG II) and potassium (K^+). The regulation of the production of aldosterone can be divided into two phases: an acute phase that takes place within minutes and reflects cholesterol transfer to mitochondrial side-chain cleavage enzyme and a chronic phase that requires several hours and reflects the increased expression of aldosterone synthase (CYP11B2) (4).

The CYP11B2 gene contains instructions for the synthesis of aldosterone synthase. This enzyme is found in the adrenal glands, which are located on top of the kidneys. Aldosterone synthase is a key enzyme in the construction of the terminal steps of aldosterone biosynthesis and a member of the cytochrome P450 family of enzymes which catalyses the terminal steps of aldosterone synthesis. Therefore, genetic mutations of the CYP11B2 gene that affect its expression may influence the development of salt-sensitive hypertension (5). Several common polymorphisms have been described in the CYP11B2 gene (6). Essential hypertension is partly genetic; one possibility is that altered control of aldosterone synthase gene expression or translation may be responsible. We compared the frequency of 2 linked polymorphisms, one in the binding site and the other in intronic conversion (IC), in groups of hypertensive and normotensive subjects (7).

Abnormalities in either of the tightly linked genes encoding the enzymes CYP11B1 or CYP11B2 can lead to important changes in arterial pressure and are responsible for several monogenicallyinherited forms of hypertension. Mutations in the CY11b2 gene or its regulatory regions could

contribute to genetic variation in receptiveness to essential hypertension (8).

The results of earlier studies are not always consistent. Differences in background characteristics of the study subjects such as ethnicity and selection criteria may be responsible for this inconsistency. In the present study, we examined the −344T/C polymorphism of the CYP11B2 gene in carefully selected normotensive subjects and patients with essential hypertension and thereby evaluated the gene expression of the CYP11B2 gene with genetic preference to essential hypertension.

Essential hypertension is a complex disorder and multi-factorial disease which involves the risks of non-genetic factors. Mostly the environmental factors that are affecting the risk genes are the foremost factors of the rise in blood pressure. Essential hypertension is partly genetic; altered control of aldosterone synthase gene expression or translation may be responsible. The risk factors of hypertension involve high sodium intake, lack of physical activity, alcohol consumption and genetically is is due to the family history of hypertension (9, 10).

Hypertension is a major global concern. It has a huge disturbing impact on the population, resulting in avoidable morbidities and mortalities. Hypertension alone is held accountable for more than 5.8 % of deaths worldwide, loss of 11.9 % years of life, adjusted life of 1.4 % and decreasing life expectancy (11).

Essential hypertension is thought to be a polygenetic disease and polymorphisms in the renin-angiotensin-aldosterone system have been studied extensively as candidate genes for cardiovascular disease including hypertension (12).

Human essential hypertension is a complex, multifactorial, and polygenic trait. There are several fundamental genes, which together contribute to between 30% and 50% of the variation in blood pressure among individuals (13).

Hypertension is one of the most common medical disorders, associated with an increased incidence of all-cause and cardiovascular disease mortality. 54% of strokes and 47% of cardiac deaths are attributed to suboptimal blood pressure control. Although a variety of anti-hypertensive medicines are present hypertension remains uncontrolled (14).

Genetic analysis of hypertension has suggested that several genetic variants increase the risk for hypertension. It is concluded that several genes rather than a single gene for heritability are involved in this complex disorder. Identifying gene variants that contribute to hypertension may not only provide a better understanding of the pathophysiology of the disease but also may illuminate the biochemical and physiological pathways that link various risk factors in hypertension. At least 51 genes/loci that affect different physiological and biochemical systems have been described for essential hypertension. Case-control design provides candidate gene studies less likely to be reproducible and less expected to include all causative genes and polymorphisms (15).

Most genetic studies of hypertension involve the association of the gene and the most associated gene is the aldosterone synthase gene (CYP11B2) (16).

The secretion of aldosterone is mainly regulated at the level of expression of the aldosterone synthase (CYP11B2) gene, with the encoding of enzymes required for the final steps of its biosynthesis. Genetic amendments can lead to inappropriate expression of CYP11B2 resulting in excessive aldosterone secretion and high blood pressure (BP), glucocorticoid suppressible hyperaldosteronism. Aldosterone is one of the main effectors of the renin-angiotensin-aldosterone system. The secretion of aldosterone is mainly regulated at the level of expression of the aldosterone synthase (CYP11B2) gene. Genetic rearrangements can lead to inappropriate expression of CYP11B2 resulting in excessive aldosterone secretion and high blood pressure (BP), glucocorticoid suppressible hyperaldosteronism. As the CYP11B2 gene is involved in the regulation of salt and water homeostasis and thereby also BP therefore other mutations at this gene locus may cause loss of function leading to salt wasting and hypotension (17).

The National Health Survey of Pakistan estimated that hypertension affects 18% of adults and 33% of adults above 45 years old. Another citation showed that 18% of people in Pakistan suffer from hypertension with every third person over the age of 40 becoming increasingly exposed to a wide range of diseases. In some remote areas like Balochistan, there is a paucity of data but the control rate is likely to get even worse (18).

From 1990 to 1994, the National Health Survey of Pakistan showed the image of hypertension in Pakistan. Hypertension was found to affect 18% of adults >15 years and 33% of adults >45 years. The present studies according to the National Health Survey of Pakistan have shown that the prevalence of Hypertension amongst adults >15 yrs was 17.9% and amongst them, only 3% had their BP under control as opposed to a prevalence of 28.7% and control of around 31% being reported from the US. Hence there is a contrast in the control of hypertension between the two countries (19. 20).

China is a culturally diverse country, and the prevalence of essential hypertension in the 56 official ethnicities varies. Essential hypertension affects approximately 20–30% of the adult population worldwide and 18.8% in China.

Despite environmental factors, genetic factors may play an important role in essential hypertension. The Kazakhs, a nomadic population dwelling in the north of Xinjiang in northwest China, have a prevalence of 18.97% for EH, which is higher than that of other ethnic populations residing in the same area (21).

From 1992 to 2000 it was estimated that a reduction in major risk factors contributed to a 50% reduction in mortality from coronary heart disease in the U.S. In 2013-14 46% of adults with hypertension did not have their blood pressure under control in the U.S. (22).

The CYP11B2 gene encodes a basic enzyme of the aldosterone synthase. Aldosterone is an independent risk factor for cardiovascular diseases and development of hypertension (23)

The renin-angiotensin-aldosterone system and its candidate genes are principally involved in the regulation of blood pressure through salt-water homeostasis. Aldosterone synthase is an important RAAS mediator which plays a major role in hypertension through the regulation of water-electrolyte balance (3).

The CYP11B2 gene provides instructions for making an enzyme called aldosterone synthase. Aldosterone synthase Is a member of the cytochrome P450 family of enzymes.

These enzymes are involved in the formation and breakdown of various molecules within cells. Aldosterone synthase helps produce a hormone called aldosterone. Aldosterone helps control blood pressure by maintaining.
Proper salt and fluid levels in the body. The aldosterone synthase enzyme is involved in a series of three chemical reactions that produce aldosterone from other molecules: the conversion of 11-deoxycorticosterone to corticosterone, the conversion of corticosterone to 18-deoxycorticosterone, and the conversion of 18-deoxycorticosterone to aldosterone.

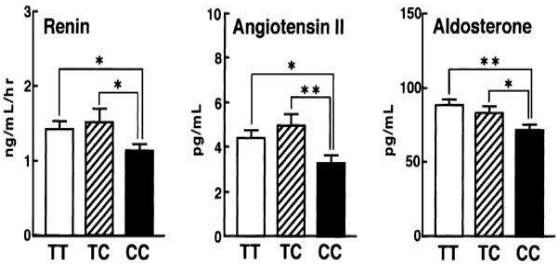

Figure 1

The secretion of aldosterone is mainly regulated at the level of expression of the aldosterone synthase (CYP11B2) gene, encoding the enzyme required for the final steps of its biosynthesis. Genetic rearrangements can lead to inappropriate expression of CYP11B2 resulting in excessive aldosterone secretion and high blood pressure. Other mutations at this gene locus may cause loss of function leading to salt wasting and hypotension. The aldosterone synthase gene (CYP1B2) locus is a candidate region involved in the development of hypertension.
Gene polymorphism is one of the reasons for hypertension. CYP11B2 is associated with hypertension and polymorphisms that occur in this gene site are the main cause of hypertension. The 344T/C polymorphismalter the site of Thymine with Cytosin. This polymorphism shows differential binding to the putative binding site (SF-I) and affects the activity of the promoter region and gene expression of the CYP11B2 gene (13).
This polymorphism has been shown to influence aldosterone levels and arterial stiffness which has been associated with hypertension (8).
Predilections to essential hypertension and cardiovascular diseases are possibly associated with gene polymorphisms of the renin-angiotensin system. The main risk factors for Hypefractions are gene polymorphisms of angiotensinogen and angiotensin-converting enzyme genes (7).
Aldosterone synthase includes the steroid 11β-hydroxylase, 18-hydroxylase and 18-oxidase activities that are required for the completion of the process of aldosterone biosynthesis. A related enzyme, 11β-hydroxylase is responsible mainly for cortisol biosynthesis, although changes in its activity can also influence biosynthesis of steroid metabolites with mineralocorticoid actions. The two closely associated genes (CYP11B2, and CYP11B1) are located in proximity on chromosome 8q22. It is now apparent from the investigation of rare monogenic forms of hypertension.CYP11B2 is considered an obvious gene to test for association with hypertension.

Three common genetic variants of the aldosterone synthase gene (CYP11B2) have been identified as possible determinants of high blood pressure in patients with essential hypertension. One is a single nucleotide polymorphism in the 5' promoter region at -344T/C that alters a putative recognition site for steroidogenic transcription factor-1 (SF-1). The other polymorphism involves intron 2 of CYP11B2, which is partly replaced by the corresponding intron of CYP11B1 gene. These two polymorphisms are in close linkage disequilibrium. The third polymorphism is a point mutation K173R in exon 3. Because the severity of hypertension determines cardiovascular risk and aldosterone is an important therapeutic target in hypertension, genotyping for the -344T/C polymorphism of the CYP11B2 gene may have prognostic as well as therapeutic potential in hypertensive patients (4).

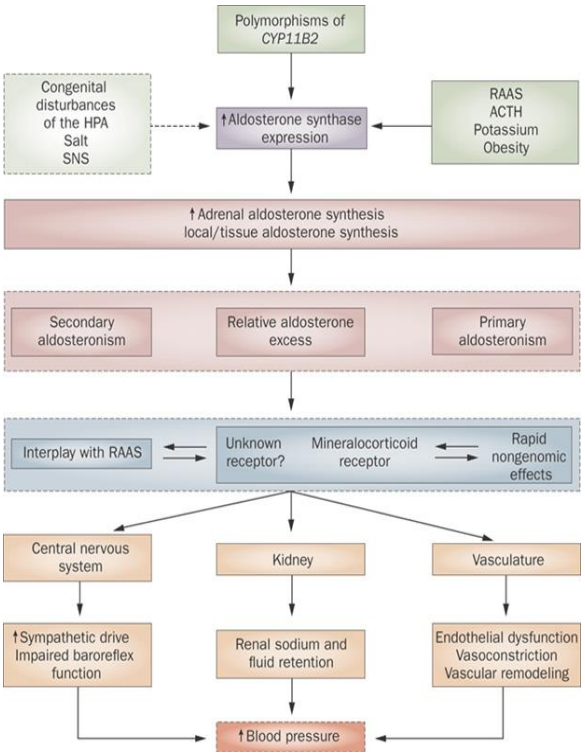

Figure 2

Mechanism of Aldosterone- mediated hypertension, Polymorphism of CYP11B2 gene, dietary salt intake. A complex interaction between genomic and non-genomic effects intercedes mineralocorticoid-induced target organ and tissue damage within the central nervous system which results in hypertension (8).

RESEARCH METHODOLOGY

Samples Collection:
The blood samples of 65 cases of hypertension and 42 controls were collected from Ganga Ram Hospital and shipped on ice to the Molecular Biology and Biotechnology Laboratory, Virtual University, 1-Davis Road campus. The samples were stored at -20C for further use.

DNA Extraction:

DNA extraction was done by using a commercial blood DNA kit (Thermo Scientific, USA) by the below-mentioned steps as per instructions.

200µL of whole blood was added to labelled Eppendorf tube tubes and 20µL of proteinase K solution, the samples were mixed gently for 15 seconds.

After mixing well, 400µL of lysis solution was added to the mixture. The suspension (Blood, proteinase K and Lysis buffer) was incubated at 56 °C for 10 minutes using shaking water.

The incubated mixture was transferred to the labelledBlistered pack DNA purification columns (available with kit).

The sample mixture (600 ul) was transferred to the labelled column; the remaining mixture was transferred again in the same way.

The columns were centrifuged at 10,000 rpm for 1 minute as per instructions. After centrifugation, collection tubes containing flowthrough solution were discarded.

Steps 5 and 6 were repeated with the remaining mixture

The DNA threads trapped in the column were purified through 500µL of working wash buffer1 was added to the column.

The tubes were centrifuged at 10,000 rpm for 1 min. The flow-through solution was discarded and the column was placed in the emptied collection tube.

The DNA was purified with 50µL of working wash buffer 2 was added to the column.

The tubes were centrifuged at 10,000 rpm for 1 min. The flow-through solution was discarded and the column was placed in an emptied collection tube.

The tubes were centrifuged into empty tubes to discard the residues at 14,000 rpm for 1 minute.

The collection tube containing the flow-through solution was discarded

The columns were transferred onto sterile 1.5µL eppendorf tubes labelled accordingly with the same number.

DNA was eluted with 200µL of elution buffer added to the column and incubated at room temperature for 2 minutes.

The tubes were centrifuged at 10,000 rpm for 1 minute.

The purification columns were discarded and the purified DNA eluted out was stored at -20C for DNA amplification.

DNA quantification:

The extracted DNA was quantified by using agarose gel electrophoresis and Nano-drop spectrophotometer c2000 (Thermo Scientific, USA). Initially, the DNA was quantified using 1% agarose gel with a DNA marker. The bands were observed through a gel documentation system.

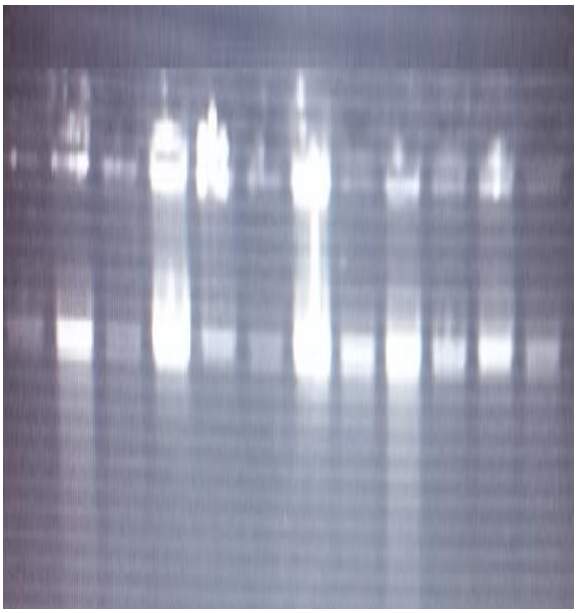

Figure 3

The extracted DNA samples were quantified through 1% agarose gel electrophoresis.

The samples were also quantified through a Nano-drop spectrophotometer c2000 (Thermo Scientific, USA). The DNA quantification and 260/280 ratio values calculated through Nano-drop have been mentioned in Table 1.

Table 1: The DNA samples quantified through Nano-drop spectrophotometer c2000 (Thermo Scientific, USA)

S. No.	Sample number	Concentration ng/ul	260/280 ratio
1	HT-D-01	21	1.81
2	HT-D-02	35	1.90
3	HT-D-03	19	1.59
4	HT-D-04	48	1.45
5	HT-D-05	12	1.65
6	HT-D-06	54	1.87
7	HT-D-07	36	1.82
8	HT-D-08	52	1.49
9	HT-D-09	32	1.74
10	HT-D-10	21	1.69
11	HT-D-11	10	1.78
12	HT-D-12	8	1.59
13	HT-D-13	25	1.82
14	HT-D-14	24	1.49
15	HT-D-15	25	1.74
16	HT-D-16	48	1.82
17	HT-D-17	52	1.49

18	HT-D-18	65	1.74
19	HT-D-19	21	1.72
20	HT-D-20	14	1.80
21	HT-D-21	45	1.59
22	HT-D-22	35	1.45
23	HT-D-23	32	1.82
24	HT-D-24	12	1.49
25	HT-D-25	22	1.74
26	HT-D-26	11	1.49
27	HT-D-27	14	1.74
28	HT-D-28	21	1.65
29	HT-D-29	25	1.82
30	HT-D-30	65	1.49
31	HT-D-31	77	1.94
32	HT-D-32	81	1.68
33	HT-D-33	21	1.77
34	HT-D-34	21	1.82
35	HT-D-35	12	1.49
36	HT-D-36	14	1.82
37	HT-D-37	200	2.21
38	HT-D-38	21	1.74
39	HT-D-39	11	1.59
40	HT-D-40	10	1.45
41	HT-D-41	9	1.72
42	HT-D-42	54	1.77
43	HT-D-43	52	1.82
44	HT-D-44	65	1.49
45	HT-D-45	25	1.74
46	HT-D-46	41	1.82
47	HT-D-47	21	1.49
48	HT-D-48	21	1.74
49	HT-D-49	22	1.69
50	HT-D-50	24	1.77
51	HT-D-51	25	1.82
52	HT-D-52	35	1.82
53	HT-D-53	25	1.49
54	HT-D-54	14	1.74
55	HT-D-55	25	1.71
56	HT-D-56	12	1.87
57	HT-D-57	9	1.80
58	HT-D-58	21	1.77
59	HT-D-59	25	1.82
60	HT-D-60	23	1.77
61	HT-D-61	21	1.81
62	HT-D-62	21	1.55
63	HT-D-63	54	1.82
64	HT-D-64	55	1.66
65	HT-D-65	65	1.65
66	HT-N-01	12	1.49
67	HT-N-02	14	1.82
68	HT-N-03	11	1.49
69	HT-N-04	21	1.74
70	HT-N-05	11	1.59
71	HT-N-06	10	1.45
72	HT-N-07	9	1.80
73	HT-N-08	54	1.77
74	HT-N-09	52	1.81
75	HT-N-10	65	1.49

76	HT-N-11	21	1.74
77	HT-N-12	41	1.72
78	HT-N-13	18	1.49
79	HT-N-14	21	1.71
80	HT-N-15	22	1.66
81	HT-N-16	24	1.77
82	HT-N-17	26	1.82
83	HT-N-18	11	1.82
84	HT-N-19	25	1.49
85	HT-N-20	14	1.74
86	HT-N-21	21	1.70
87	HT-N-22	12	1.80
88	HT-N-23	9	1.80
89	HT-N-24	21	1.77
90	HT-N-25	25	1.82
91	HT-N-26	23	1.77
92	HT-N-27	74	1.81
93	HT-N-28	21	1.55
94	HT-N-29	25	1.82
95	HT-N-30	55	1.66
96	HT-N-31	31	1.65
97	HT-N-32	21	1.71
98	HT-N-33	25	1.82
99	HT-N-34	23	1.77
100	HT-N-35	21	1.89
101	HT-N-36	21	1.55
102	HT-N-37	54	1.80
103	HT-N-38	45	1.59
104	HT-N-39	14	1.64
105	HT-N-40	18	1.79

Primers used for the amplification of the target gene:
The following pair of reported primers (Setyawati*et al.*,2016) was used for the DNA amplification of the target gene Pan troglodytes cytochrome P450 family 11 subfamily B member 2 (CYP11B2)by using Bio-Rad PCR machine T-100.
Forward primer
5'-CAGGAGGGATGAGCAGGCAGAGCACAG-3'
Reverse primer
5'CTCAACCCAGGAACCTGCTCTGGAAACATA-3'

PCR Amplification:
The target gene was amplified by using the reported primer for screening of −344T/C polymorphism of the CYP11B2 gene. The PCR reaction mixture included the 20-50 ng/genomic DNA, 10 pM forward and reverse primers, 2.5 mM MgCl2, buffer (*ammonium* sulfate $(NH_4)_2SO_4$), Deoxynucleotide triphosphate DNTPs (2.5mM), Taq DNA polymerase enzyme (5 U), and deionized water in a 0.5 ml PCR tube. The PCR was performed in a Bio-Rad T00 PCR machine with the following PCR conditions shown in Table 3.2. The PCR profile included initial denaturation at 95°C for 5 minutes followed by 35 cycles of denaturing at 95°C for 60 s, annealing at 67°C for 60 s, and polymerization at 72°C for 120 s and final extension at 72°C for 10 minutes and store the reaction at 4°C

Restriction Fragment Length Polymorphism-RFLP:
−344T/C polymorphism of the CYP11B2 gene was determined by the analysis of restriction fragment length polymorphism (RFLP). The PCR products were digested with with*HaeIII*Restriction enzyme and were subjected to electrophoresis on 2.0% agarose gels. *HaeIII* digestion of the 538 bp PCR product yields 274, 138 and 126 bp fragments. The existence of −344C creates an additional recognition site of *HaeIII* resulting in indigestion of the 274 bp fragment into 203 and 71 bp fragments. Fragments of 274 bp (T allele) and 203 and 71 bp (C allele) were detected.

Table 2: Composition of PCR reaction mixture

Reagents	Concentration	Volume
DNA	20 to 30 ng/µl	1.0 µl
Forward Primer	10 pm	1.0 µl
Reverse Primer	10 pM	1.0 µl
dNTPs	2.5 mM	2.5 µl
Taq Polymerase	5 U	0.5 µl

$MgCl_2$	2.5 mM	2.5 µl
Buffer		2.5 µl
Water	(DEPC)	14.0 µl
Total		25 µl

Table 3: The PCR Condition

Steps	Temperature	Duration
Initial Denaturation	95ºC	5 min
1.Denaturation	95ºC	60 sec
2. Annealing	67ºC	60 sec
3. Extension	72ºC	120 sec
Repeat 1-3 steps for 35 cycles.		
Final Extension	72ºC	10 min

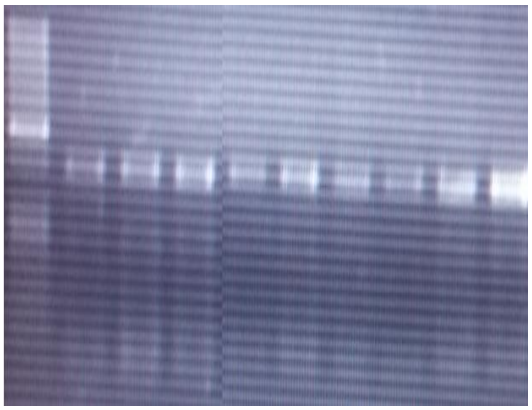

Figure 4 PCR product of CYP11B2 gene

Restriction Fragment Length Polymorphism-RFLP: −344T/C polymorphism of the CYP11B2 gene was determined by the analysis of restriction fragment length polymorphism (RFLP). The PCR products were digested with with*HaeIII*Restriction enzyme and were subjected to electrophoresis on 2.0% agarose gels. *HaeIII* digestion of the 538 bp PCR product yields 274, 138 and 126 bp fragments. The existence of −344C creates an additional recognition site of *HaeIII* resulting in indigestion of the 274 bp fragment into 203 and 71 bp fragments. Fragments of 274 bp (T allele) and 203 and 71 bp (C allele) were detected**.**

Results

In the present research work, a total of 41 men and 24 women were in the case group, while 32 men and 9 women were in the control group without a hypertension history. There was no significant difference in the distribution of age, body weight, body mass index (BMI), body height, systolic blood pressure, and diastolic blood pressure between the control and case groups.

Allelic frequency distribution in cases and controls is shown in Table 1, while the frequency distribution of the TT, TC and CCgenotypes in the case and control groups is shown in Table 3.2. Controls were with a higher frequency of TT genotype whereas patients were with both CT and CC genotypes shown in Table 3.2. The C allele was not observed in the control samples and more frequently observed in the diseased patients was determined to be a risk factor for hypertension with < 0.01.

Table 4.1: Allelic frequency distribution in cases and controls

Gene	Allele	Patients (N/Frequency)	Controls (N/Frequency)	HWE (p-value)	X^2
CYP11B2	C	51 (0.784)	0 (0.000)	1.000000	60.379150
	T	14 (0.216)	42 (1.000)		

CYP11B2: HWE: Hardy Weinberg equilibrium, n= number, X^2= chi square value, Significance level: p < 0.01

Table 4.2: Genotype frequencies distribution in cases and controls

Gene	Genotype	Patients N (Frequency)	Controls N (Frequency)	X^2
VDR	CC	11 (0.169)	0 (0.000)	57.777779
	CT	40 (0.615)	0 (0.000)	
	TT	14 (0.216)	42 (1.000)	

CYP11B2: n= number, X^2= chi-square value, Significance level: p < 0.01

Discussion

In this study, the frequency of the TT genotype was higher in the control than in the cases. The TC and CC genotypes were higher in cases than in the controls, and they were risk factors for hypertension. This result coincided with studies carried out in Japanese and Chinese populations. In this study, we have found higher aldosterone levels in individuals with the CC genotype compared to individuals with the TC+ TT genotype and in individuals with the TC+ CC genotype compared to individuals with the genotype. However, this difference was not significantly significant. The TC and CC genotypes were found more frequently in cases than in the controls, but the aldosterone levels in the two groups were not significantly different.

K Tsukada et al investigated the association between the 344T/C polymorphism in the promoter region of the human CYP11B2 gene and the development of hypertension. The results indicate that the genotype distribution differs between hypertensive and normotensive groups and that the 344C allele is associated with a genetic predisposition to develop EH. 344C allele of CYP11B2 gene polymorphism was associated with left ventricular mass in Finnish young adults without clinical heart diseases. They have also reported that the 344C allele was associated with higher systolic blood pressure levels in Finnish men.

If we observe the study by Kato et al, they have performed a case-control study in which they failed to find any relation between the gene polymorphisms of the renin-angiotensin-aldosterone system and essential hypertension. Discordance between the results of the study by Kato et al with our results may be featured to the influence of environmental factors. The quality and quantity of stress, which contribute to the development of hypertension, are supposed to be different in people of different countries.

Concerning the plasma aldosterone levels, Pojoga et al have shown that the CC genotype was associated with increased plasma aldosterone in French patients with EH, while Brand et al have reported that plasma aldosterone was not affected by the genotypes of 344T/C polymorphism in German normotensive subjects.

Davies et al have reported that urinary excretion of aldosterone is less in the CC genotype than in the TC and TT genotypes. That's why it is observed that the CC genotype may be related to the development of low renin hypertension however, the mechanism is not supposed to be brought about by the increased transcription of the aldosterone synthetase gene.

Declarations

Data Availability statement

All data generated or analyzed during the study are included in the manuscript.

Ethics approval and consent to participate

Approved by the department concerned. (IRBEC-TCHLL-0093/23)

Consent for publication

Approved

Funding

Not applicable

Conflict of interest

The authors declared the absence of a conflict of interest.

Author Contribution

SEEMAB MAZHAR
Coordination of collaborative efforts.
Study Design, Review of Literature.
ABDUL WAJID (Assistant Professor)
Conception of Study, Development of Research Methodology Design, Study Design, Review of manuscript, final approval of manuscript.
Conception of Study, Final approval of manuscript.
UZMA FIRDOUS (Assistant Professor)
Manuscript revisions, critical input.
Manuscript drafting.

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